Published in final edited form as:

Psychopharmacology (Berl). 2016 December ; 233(23-24): 3905–3919. doi:10.1007/s00213-016-4423-6.

Oxytocin modulates hemodynamic responses to monetary incentives in humans

Brian J. Mickey^{1,2,3}, Joseph Heffernan², Curtis Heisel², Marta Peciña^{1,2}, David T. Hsu^{1,2,4}, Jon-Kar Zubieta^{1,2,3}, and Tiffany M. Love^{1,2,3}

¹Department of Psychiatry, University of Michigan Medical School, Ann Arbor, USA

²Molecular and Behavioral Neuroscience Institute, University of Michigan, Ann Arbor, USA

³Department of Psychiatry, University of Utah School of Medicine, Salt Lake City, USA

⁴Department of Psychiatry, Stony Brook School of Medicine, Stony Brook, New York, USA

Abstract

Oxytocin is a neuropeptide widely recognized for its role in regulating social and reproductive behavior. Increasing evidence from animal models suggests that oxytocin also modulates reward circuitry in non-social contexts, but evidence in humans is lacking. Here we examined the effects of oxytocin administration on reward circuit function in 18 healthy men as they performed a monetary incentive task. The blood oxygenation level dependent (BOLD) signal was measured using functional magnetic resonance imaging in the context of a randomized, double-blind, placebo-controlled, crossover trial of intranasal oxytocin. We found that oxytocin increases the BOLD signal in the midbrain (substantia nigra and ventral tegmental area) during the late phase of the hemodynamic response to incentive stimuli. Oxytocin's effects on midbrain responses correlated positively with its effects on positive emotional state. We did not detect an effect of oxytocin on responses in the nucleus accumbens. Whole-brain analyses revealed that oxytocin attenuated medial prefrontal cortical deactivation specifically during anticipation of loss. Our findings demonstrate that intranasal administration of oxytocin modulates human midbrain and medial prefrontal function during motivated behavior. These findings suggest that endogenous oxytocin is a neurochemical mediator of reward behaviors in humans – even in a non-social context – and that the oxytocinergic system is a potential target of pharmacotherapy for psychiatric disorders that involve dysfunction of reward circuitry.

Introduction

Oxytocin is a centrally active neuropeptide best known for its roles in social and reproductive behavior. The mechanisms by which oxytocin modulates these behaviors are not completely understood. Current models include the approach-avoidance model (Kemp and Guastella 2011) and the social salience model (Bartz et al. 2011). It has also been

Correspondence: Tiffany Love, PhD, tiffany.love@hsc.utah.edu, Phone: 801-587-0160, Fax: 801-581-7109.

Conflict of interest: During the 5-year period prior to submission, BJM served as a consultant to Alkermes, Inc., for work unrelated to this manuscript.

ClinicalTrials.gov registration NCT01722071 (<http://clinicaltrials.gov/ct2/show/NCT01722071>)

suggested that oxytocin facilitates social interaction by modulating activity within reward networks (Gordon et al. 2011; Love 2014). Oxytocin receptors are widely distributed within reward-sensitive brain regions including the ventral tegmental area (VTA), nucleus accumbens (NAc), and prefrontal cortex (PFC) (animals: Adan et al. 1995; humans: Boccia et al. 2013; Dumais et al. 2013; Loup et al. 1991). Oxytocin neurons project from the paraventricular and supraoptic nuclei of the hypothalamus to the NAc and VTA, where they have been shown to regulate dopamine neurotransmission (animals: Melis et al. 2007; Melis et al. 2009). As dopamine is thought to modulate attributions of salience towards reward-predicting stimuli, it has been suggested that oxytocin, through its interface with mesoaccumbal dopamine systems, may influence the salience of incentive cues which could ultimately affect an individual's drive towards significant stimuli (Becker and Taylor 2008).

Indeed, oxytocin alters reward-driven behavior associated with a variety of incentives in experimental animals via modulation of mesoaccumbal dopamine release. Oxytocin infusions into the NAc and VTA enhance dopamine release in association with increased performance of socially motivated behaviors such as pup grooming, pair bonding, and sexual behaviors (Gordon et al. 2011). Oxytocin directly administered into the NAc can also attenuate reward-related behaviors associated with drugs of abuse, including drug self-administration and behavioral sensitization, and interfere with drug-induced dopamine release in rats (Sarnyai and Kovács 2014).

Although an extensive animal literature provides evidence that oxytocin shapes neural and behavioral responses to reward, whether these findings generalize to humans is still being explored. Several recent human studies have suggested that oxytocin administration can influence neural responses to social stimuli, including faces (Groppe et al. 2013; Scheele et al. 2013), social touch (Scheele et al. 2014), and cooperative interactions (Rilling et al. 2014). Lee and colleagues recently reported that intranasal oxytocin enhanced *behavioral* responses to monetary reward in abstinent cocaine users (Lee et al. 2014). To our knowledge, the effects of oxytocin on *neural* responses to non-social rewards have been examined in just one study of trauma-exposed police officers with and without PTSD (Nawijn et al. 2016). That study suggested that oxytocin enhanced responses to monetary incentives in the putamen, insula, and anterior cingulate cortex, but no oxytocin effects were detected in the mesoaccumbal pathway, and no effects survived whole-brain correction for multiple comparisons (Nawijn et al. 2016).

In the present study, we tested the hypothesis that exogenously administered oxytocin influences mesoaccumbal responses to monetary incentive stimuli in healthy humans. We focused on the mesoaccumbal pathway because midbrain dopaminergic projections to the nucleus accumbens are known to drive reward behavior in humans and animals and these pathways are subject to oxytocin modulation. We approached this question using functional magnetic resonance imaging in the context of a randomized, double-blind, placebo-controlled, crossover trial of intranasal oxytocin. We used an fMRI task that robustly activates the mesoaccumbal reward pathway, the Monetary Incentive Delay (MID) task (e.g. Cooper and Knutson 2008; Knutson et al. 2004; Knutson et al. 2001). We predicted that oxytocin administration would increase activity in midbrain and nucleus accumbens. There is evidence to suggest that oxytocinergic effects may be salience (e.g. Wittfoth-Schardt et al.

2012) or valence-dependent (e.g. Domes et al. 2013b; Gamer and Büchel 2012; Gamer et al. 2010; Kis et al. 2013). As such, we chose to utilize a version of the MID task, which varies incentives along both of these dimensions. Based on suggestions that personality traits may moderate the behavioral effects of oxytocin (Bartz et al. 2011), an additional exploratory aim was to examine how personality traits moderate the neural effects of oxytocin.

Materials and Methods

Study design

All procedures were approved by the University of Michigan Institutional Review Board and performed at the University of Michigan Medical Center in 2013–14. Protocols were registered on [ClinicalTrials.gov](https://clinicaltrials.gov) (identifier: NCT01722071). We used a double-blind, randomized, placebo-controlled, crossover design. Each participant was scanned twice: once after oxytocin, and once after placebo. Scans were performed on different days 1–2 weeks apart, and the order of scans was randomized and counterbalanced across participants. Randomization was achieved with a computer-generated random allocation sequence (SAS software, Cary, NC) without blocking, and allocation was concealed prior to scanning to prevent selection bias. Participants and study personnel who interacted with participants were thus blinded to drug assignment (oxytocin versus placebo) until the completion of the study; only the research pharmacy personnel who prepared the drugs were not blinded.

Participants

Twenty healthy men were recruited by advertisement and provided written informed consent. The sample size of 20 was chosen based on power analysis which indicated 90% power to detect a standardized effect size > 0.8 , allowing for 10% attrition and 5% type I error. Participants were screened with the Mini International Neuropsychiatric Inventory, version 6.0.0 by a PhD level study member who was supervised by a psychiatrist (Sheehan et al. 1998). Eligible subjects were right-handed men, age 20–35 y, with no current medications and no history of neurological, psychiatric, or substance use disorder (including tobacco). One subject was excluded before scanning due to a history of depression. Another subject was later excluded from data analysis due to excessive movement during functional MRI scanning, leaving 18 subjects available for analysis.

Drug

Oxytocin and matched placebo preparations were obtained from PharmaWorld (Zurich, Switzerland). The oxytocin preparation was identical to placebo except for the addition of 4 international units of oxytocin per 0.1 ml (Syntocinon®, Novartis, Basel, Switzerland).

Prior to drug administration, study staff confirmed that breathing was unobstructed and that participants were not experiencing congestion. Participants were then given verbal instructions on how to administer the drug and the proper nasal delivery action was demonstrated by study staff. While being monitored by study staff to ensure compliance, participants were then asked to self-administer the intranasal drug as follows: After confirming the intranasal device was primed by completing a test spray, participants, while keeping their head upright, were instructed to insert the intranasal device into their nose

(≤ 0.5 inch) and to aim away from the midline. After the device was in place, participants were asked to depress the pump and to gently breathe in. Participants were asked to alternate sprays between nostrils. Participants self-administered oxytocin and placebo intra-nasally by applying three 0.1-ml puffs per nostril (24 units of oxytocin in total).

The reward task in the MRI scanner began 49 ± 18 minutes (mean $\pm$ SD) after administration of oxytocin or placebo. This timing was chosen based on previous functional MRI studies that investigated the neural effects of oxytocin (Marsh et al. 2010; Riem et al. 2011a; Riem et al. 2011b; Rimmelle et al. 2009) and it aligns with previous reports of neuropeptide elevations in cerebrospinal fluid following intranasal administration in monkeys and humans (Born et al. 2002; Dal Monte et al. 2014).

Following each scan, participants were asked to guess whether they had received oxytocin or placebo. To examine whether patient blinding was successful, we examined these data using an exact binomial test in R. Results indicated that participants were not able to successfully detect above chance which drug they received during each scan: 9 out of the 18 participants correctly guessed which drug they had received during their first scan ($p=1.0$) and 7 out of the 18 correctly guessed during their second scan ($p=0.48$).

Questionnaires

Emotional states were measured with the Positive and Negative Affect Schedule [PANAS (Watson and Clark 1994)], the Spielberger State-Trait Anxiety Inventory [STAI (Spielberger et al. 1983)], and the Brief Fear of Negative Evaluation (BFNE) scale (see Supplementary Materials for a complete description of each measure). These questionnaires were completed just prior to the MRI scan, which began 30 ± 6 minutes (mean $\pm$ SD) after oxytocin or placebo administration.

Personality traits were characterized with the Revised NEO Personality Inventory (NEO PI-R), which reliably measures five major domains: Neuroticism, Extraversion, Openness, Agreeableness, and Conscientiousness (Costa and McCrae 1992).

Reward task

To examine the effects of oxytocin administration on neural responses to non-social incentive stimuli, we used a modified version of the monetary incentive delay task (MID, Figure 1, Cooper and Knutson 2008). During this task, participants were instructed they would play a game where they had the chance to win money. Participants were told that on the beginning of each trial they would see one of five cues (see *Trial conditions* below) which would signal the trial type and the amount of money at stake, then after a delay, a solid white target box would briefly appear. Participants were informed that their goal was to press a button as quickly as possible as soon as the target box appeared on the screen. Participants were instructed to try to hit the target regardless of trial condition. The presentation time for the target box was adjusted dynamically based on a subject's reaction time to maintain an average target hit rate near 60%. This was done to assure consistency of success across participants and between sessions. Participants were not told in advance that the target box duration would be adaptively adjusted based on their performance. At the end of each trial, the participant received feedback about the outcome of the trial. The five trial

conditions were each presented 9 times during each session (27 times each over 3 sessions) in a pre-defined pseudorandom order. The order of stimuli was identical across subjects and drug treatments.

Trial conditions

In this version of the MID task, trials varied across two dimensions: salience (high and low) and valence (reward and loss). As such, there were 5 trial conditions in total: high-salience reward, low-salience reward, high-salience loss, low-salience loss, or neutral (Figure 1). This task was chosen over other reward tasks based on evidence that oxytocinergic effects may be salience-dependent (Wittfoth-Schardt et al. 2012) or valence-dependent (Domes et al. 2013a; Gamer and Büchel 2012; Gamer et al. 2010; Kis et al. 2013). Valence was manipulated by varying the type of incentive at stake (positive-valence = reward; negative-valence = loss); whereas salience was manipulated by varying the certainty of the outcome (see Cooper and Knutson 2008). In high-salience trials (indicated by '?'), participants were instructed that the button must be pressed when the target was on-screen in order to win or avoid losing money. If a participant successfully responded to the target during high-salience trials, the subject either gained \$3 (reward trials) or avoided losing \$3 (loss trials). In low-salience trials, subjects were told that while they should respond to the target box, whether they hit the target had no impact on the amount of money they won or lost for that trial. In other words, the outcome was certain during low-salience trials: the subject always won \$3 (reward trials) or lost \$3 (loss trials). On neutral trials, no money was at stake, but participants were instructed to try to hit the target nonetheless.

Image acquisition

Whole brain functional images were acquired on a Philips Ingenia 3-T system (Best, Netherlands) using a T2*-sensitive echo planar imaging sequence (39 axial slices, slice thickness 3.5 mm, echo time 28 ms, repetition time 2 s, flip angle 90°, no slice gap, field of view 240 mm², acquisition matrix 64 × 64, ascending acquisition). Three sessions of 295 volumes each were acquired. The behavioral task was implemented using E-prime software with the Integrated Functional Imaging System (IFIS) toolbox (Psychology Software Tools, Sharpsburg PA). Stimuli were displayed on a computer monitor located at the rear of the scanner and viewed through a mirror attached to the head coil. A high-resolution structural image was obtained for anatomic normalization using a T1-weighted, inversion-recovery prepared gradient echo (MPRAGE) sequence (220 slices, slice thickness 1 mm, echo time 4.6 ms, repetition time 9.8 ms, flip angle 8°, field of view 240 mm²). Cardiac and respiratory activity were measured continuously during functional imaging using an MRI-compatible pulse oximeter and thoracic belt transducer (Philips, Best, Netherlands), respectively, and heart and respiratory rates were calculated with custom MATLAB routines (MathWorks, Natick, MA). Unfortunately, due to equipment failure, cardiac and respiratory data were only available in 15 subjects.

Image processing

Functional images were preprocessed using FSL (version 5.0.2.2, <http://www.fmrib.ox.ac.uk>), SPM8 (<http://www.fil.ion.ucl.ac.uk>), and MATLAB software (version 7.14.0.739). Images were slice-time corrected (middle reference slice) and realigned to the

mean of the time series data (SPM8). All datasets were reviewed for motion and sessions were excluded if head motion between consecutive frames was greater than 3 mm in any direction. The high resolution T1-weighted image was co-registered to the functional images using a boundary-based registration algorithm (FSL FLIRT), segmented into tissue probability maps (FSL FAST), and normalized to Montreal Neurological Institute (MNI) space (FSL FNIRT). Functional images were normalized with FSL FNIRT deformation parameters, resampled to isotropic 2-mm voxels, and smoothed with a 6-mm Gaussian kernel (SPM8).

Region-of-interest analysis

Previous fMRI studies have demonstrated that anticipation of reward recruits regions along the mesolimbic pathway including the NAc (e.g. Carter et al. 2009; Cooper and Knutson 2008; Knutson et al. 2004; Knutson et al. 2001) and VTA (Carter et al. 2009; Krebs et al. 2011). Further, recent fMRI studies indicate oxytocin administration can alter neural responses within the NAc and VTA in response to salient social stimuli such as faces (Groppe et al. 2013; Scheele et al. 2013), sexual stimuli (Gregory et al. 2015), and in response to cooperative interactions (Rilling et al. 2014). As such, we chose to specifically examine the possibility that oxytocin would modulate the hemodynamic response function in the mesoaccumbal pathway. To do so, we performed volume-of-interest (VOI) analyses for the midbrain [combined ventral tegmental area (VTA) and substantia nigra pars compacta (SN)] and left and right nucleus accumbens (NAc) (Figure 2). Left and right NAc VOIs were anatomically defined using the Harvard-Oxford subcortical structural atlas in FSL (version 5.0.2.2). The midbrain VOI was anatomically defined based on a high-resolution study of the VTA/SN (Eapen et al. 2011) and drawn on a Montreal Neurological Institute canonical brain (Wager lab, University of Colorado; http://brainstemwiki.colorado.edu/lib/exe/fetch.php/avg152_vta.nii.zip; accessed May 8, 2015). The BOLD signal time course was extracted and averaged across all voxels within the VOI. The signal change (relative to the regional mean signal) was linearly de-trended and high-pass filtered with a 100-s cutoff. Signal time courses for the initial 20-s period (11 time points) following cue onset were averaged across trials (for each participant and each task condition) and normalized to z-scores within-subjects. The effect of oxytocin versus placebo treatment on the BOLD signal time course was evaluated with a linear mixed model (fixed effects: time, treatment, condition, time $\times$ treatment, time $\times$ condition, treatment $\times$ condition, time $\times$ treatment $\times$ condition; random effect: subject) implemented with R software (version 3.0.2) and the lme4 package (version 1.0–5). Additional linear mixed models were used to examine effects per condition (Table S1). Because brainstem signals can be sensitive to cardiac and respiratory artifacts, we used paired t tests to determine whether heart and respiratory rates were influenced by oxytocin versus placebo (Table 1, Supplementary Material).

Whole-brain analysis

As oxytocin administration has been observed to influence regions outside of the mesoaccumbal pathway, we performed a planned, voxel-wise, analyses were performed in order to test for oxytocin effects outside the mesoaccumbal pathway. A two-level procedure was used. At the first level, a general linear model (SPM8) was constructed for each

participant including parameters of interest corresponding to the anticipation of each condition type, modeled as impulse functions time-locked to the presentation of each of the five anticipatory cues (i.e. cue onset). We also included the following regressors of no interest: 3 position coordinates, 3 angular coordinates, a first-order parametric modulator for each condition's regressor weighted with that trial's reaction time in order to account for potential trial-by-trial differences in motor performance (Cooper and Knutson 2008), and the top 5 principal components of the BOLD time series from white matter and CSF masks to remove signals arising from respiratory and cardiac cycles [aCompCor (Behzadi et al. 2007)]. Scanner drift and other low-frequency noise was filtered out using 100-s high-pass threshold. The AR(1) model in SPM8 was used to account for temporal autocorrelation. Four contrasts of interest were investigated - high-salience reward, low-salience reward, high-salience loss, and low-salience loss (all relative to neutral trials) – to characterize hemodynamic responses during the anticipatory phase of the task. The effect of oxytocin treatment versus placebo was tested in second-level analyses using SPM8 paired t tests with cluster-based correction for multiple comparisons.

Results

Behavioral effects of oxytocin

Eighteen healthy men (age, 22 ± 2 years, mean $\pm$ SD) completed the study and had usable data. We found no significant main effects of oxytocin treatment on emotional state as measured with the Positive and Negative Affect Schedule (PANAS), State-Trait Anxiety Inventory (STAI), and Brief Fear of Negative Evaluation (BFNE) scale (Table 1).

The monetary incentive delay task was performed in the MRI scanner. Hit rates were not affected by oxytocin treatment (Table 1). Repeated-measures ANOVA demonstrated significant main effects of task condition on hit rate ($F_{4,68} = 28.0$, $p < 0.001$). While the algorithm used to adjust task difficulty ensures an average hit rate of ~60% over the course of the experiment, it does not control for each task condition's hit rate separately. As such, between condition differences in hit rate are still possible. This particular observation, that hit rates tend to be higher under incentivized conditions, has been noted by others investigating the MID task (e.g. Cooper and Knutson 2008; Helfinstein et al. 2013). There was no effect of oxytocin on hit rate (main effect of treatment, $F_{1,17} = 1.18$, $p = 0.29$; treatment $\times$ condition interaction, $F_{4,68} = 0.54$, $p = 0.71$). Hit rate did not vary as a function of scan order (paired t-test, $t=0.26$, $df=17$, $p=0.80$). Reaction times were slightly but significantly slower following oxytocin, especially with the high-salience loss condition (Table 1). Repeated-measures ANOVA demonstrated an expected main effect of task condition on reaction time ($F_{4,68} = 15.0$, $p < 0.001$) and a significant main effect of oxytocin treatment (main effect of treatment, $F_{1,17} = 5.88$, $p = 0.03$), but no treatment $\times$ condition interaction ($F_{4,68} = 0.52$, $p = 0.72$). We saw a tendency for overall reaction times to be lower during the second scan (260ms) compared to the first scan (276ms; $t=3.06$, $df=17$, $p=0.01$). However, the results of the repeated-measures ANOVA were unchanged when the order of oxytocin/placebo administration was included as a covariate.

Midbrain effects of oxytocin

We measured BOLD signal changes in two primary regions of interest, the midbrain (combined VTA and SN) and bilateral NAc. Figure 3 shows the mean BOLD signal time course in the midbrain, time-locked to the onset of incentive cues. As expected, hemodynamic responses were greater for the more highly incentivized conditions (high-salience reward and high-salience loss conditions, Figure 3B and 3D). Compared to placebo, oxytocin treatment caused an increase in BOLD signal during the late phase of the hemodynamic response (~12 seconds after stimulus onset) across task conditions (Figure 3). We fit two linear mixed models to the hemodynamic response time course. The 4-level model adhered to the original task design, which included neutral as a reference condition. In that model, task condition was modeled as a factor with 4 levels: low-salience reward – neutral, high-salience reward – neutral, low-salience loss – neutral, and high-salience loss – neutral. To explore the possibility that oxytocin administration may potentially influence the time course of the neutral condition, we fit a second 5-level model which considered neutral as a separate condition. In that model, task condition was modeled as a factor with 5 levels: low-salience reward, high-salience reward, low-salience loss, high-salience loss, and neutral. The 5-level model revealed significant treatment-by-time and treatment-by-condition effects whereas the 4-level model showed a significant treatment-by-time effect and no significant treatment-by-condition effect (Table 2). The treatment-by-time effect indicated that oxytocin changed the BOLD time course. The treatment-by-condition effect indicated that the oxytocin's effects varied across task conditions. The results were essentially unchanged when the order of oxytocin/placebo administration was included as a covariate.

To further investigate this interaction of oxytocin treatment, time, and task condition, we used post hoc models to analyze the midbrain BOLD response for each task condition (Table S1). Significant main effects of treatment were observed for low-salience reward and neutral conditions. Significant treatment-by-time effects were observed only under the high-salience reward condition. Examination of raw BOLD time course data (without subtracting the neutral condition) suggested that oxytocin increased the late BOLD response (12-s lag) under reward conditions, decreased the late BOLD response under the neutral condition, and had no effect under loss conditions (Figure S1). To address the question of valence or salience specificity, we modeled the response at the 12-second time point with the following predictors: oxytocin treatment, valence (reward vs loss), salience (high vs low), and all 2- and 3-way interactions. Significant effects were found for treatment ($p = 0.003$), salience ($p = 0.02$), and treatment-by-valence ($p = 0.04$); all other terms were non-significant ($p > 0.2$). These observations suggest that oxytocin's effects on midbrain BOLD response may be valence specific, with greater effects under reward conditions than under loss conditions with an opposite, independent effect on midbrain responses to neutral stimuli.

The effects of oxytocin on the late phase of the midbrain hemodynamic response suggested that oxytocin could be altering responses to later trial events -- especially the visual feedback regarding the outcome of the trial -- rather than altering anticipatory responses to incentive stimuli per se. To test that possibility, we performed additional analyses in which the hemodynamic response time course was time-locked to the onset of visual feedback. Linear mixed model fits to the hemodynamic response time course in the midbrain confirmed no

significant main effects or interactions with oxytocin treatment (data not shown). These results indicate that the increase in the BOLD signal by oxytocin, despite being delayed by ~12 seconds relative to stimulus onset, was specific to incentive anticipation.

Accumbal effects of oxytocin

In contrast to the midbrain findings, hemodynamic responses in the NAc were not affected by oxytocin (Figure 4). Linear mixed model fits to the hemodynamic response time course in left and right NAc confirmed no significant main effects or interactions with oxytocin treatment (data not shown).

Prefrontal effects of oxytocin

We tested for effects of oxytocin in other brain regions using SPM8 with the built-in canonical hemodynamic response function. Oxytocin effects were found for the high-salience loss condition within a large cluster in the medial PFC ($P_{FWE} = 10^{-7}$, Table 3, Figure 5A–C). Examination of BOLD signal extracted from this cluster showed that, relative to placebo, oxytocin attenuated the negative BOLD response to high-salience loss stimuli (Figure 5D). Two smaller clusters in the left lateral prefrontal cortex showed a similar pattern but did not survive correction for multiple comparisons (Table 3). No significant effects of oxytocin were found for low-salience reward, high-salience reward, or low-salience loss conditions (whole-brain corrected $P_{FWE} > 0.05$).

The findings that medial PFC responses, late midbrain responses, and reaction times were all influenced by oxytocin treatment under the high-salience loss condition suggested that these three effects might be related. To examine this possibility, we tested for association between oxytocin-induced change in medial PFC peak hemodynamic response ($t = 6$ s; see Figure 5D) and oxytocin-induced change in mean reaction time during high-salience loss trials. We found no significant correlation across participants ($\rho = 0.04$, $p = 0.88$, $n = 18$, Spearman correlation). Similarly, we found no association between change in medial PFC peak response and change in the midbrain late peak response during high-salience loss trials ($\rho = 0.20$, $p = 0.41$, $n = 18$, Spearman correlation). Finally, there was no significant association between change in the midbrain late peak response and change in mean reaction time during high-salience loss trials ($\rho = -0.11$, $p = 0.66$, $n = 18$, Spearman correlation). Thus, we found no evidence that oxytocin's effects on reaction time, midbrain response, and medial PFC response are interdependent.

Emotion and personality correlates

Although we found no main effect of oxytocin treatment on questionnaire-measured emotional states (PANAS; see Table 1) oxytocin-induced change in emotional state varied considerably across participants. If the neural effects of oxytocin that we observed in the midbrain and medial PFC underlie oxytocin effects on subjective state, then one would expect to observe a correlation between neural and subjective state. To test this possibility, we first calculated the effect of oxytocin on midbrain response for each subject at each time point, irrespective of task condition using paired t-tests. We confirmed that the strongest effects of oxytocin were observed 12 seconds after cue onset (Figure 6A). Then, we calculated the effect of oxytocin on emotional state (PANAS positive and negative scales)

for each subject using paired t-tests. Finally, we tested, at each time point, whether change in BOLD response, irrespective of task condition, correlated with change in emotional state. We found significant Pearson correlations between change in the PANAS positive scale and change in the midbrain BOLD response at 6 and 12 seconds ($r=0.47$, $p=0.049$; $r=0.55$, $p=0.02$, respectively), corresponding to early and late hemodynamic response peaks (Figure 6B–D). No significant correlations were found with the PANAS negative scale (all $p > 0.05$). A similar analysis of medial PFC responses to high-salience loss revealed no significant correlations with PANAS positive or negative scales (all $p > 0.05$). These findings link oxytocin's effects on positive emotions with its effects on neural responses in the midbrain but not the medial PFC. Given the large number of correlation tests performed in these exploratory analyses (i.e., 11 time points), these findings should be considered provisional.

Finally, we performed an exploratory analysis of personality traits as potential moderators of the neural effects of oxytocin. Using time-course analyses similar to those described above for PANAS changes, we found no significant correlations between NEO-PI-R domain scores versus oxytocin-induced change in BOLD response in the midbrain or medial PFC (all $p > 0.05$).

Discussion

Although oxytocin is renowned for its ability to shape social behaviors, increasing evidence supports the idea that oxytocin may more generally influence neural responses to multiple forms of reward through its modulation of the mesoaccumbal and mesocortical pathway (Love 2014). Here, we provide the first evidence in humans that oxytocin administration affects mesoaccumbal responses to non-social incentives. We observed that intranasal oxytocin prolonged BOLD responses during the anticipation of monetary incentives within the midbrain but not the NAc. Furthermore, we found that oxytocin attenuated medial PFC deactivation specifically during anticipation of monetary loss. Finally, we observed that oxytocin's effects on positive emotional state correlated with its effects on midbrain responses, indicating a link between neural activity and subjective experience.

Our study has several limitations. First, the within-subject design is well-suited to detecting effects of oxytocin, but with a sample size of 18 subjects, tests for between-subjects effects, for example correlations with personality, are not well-powered to detect small-to-moderate effects. Second, the intranasal oxytocin administration protocol we used was convenient and well-tolerated, but this route is subject to between-subject variation in bioavailability and pharmacokinetics. Newer devices may reduce between-subject and between-study variation in oxytocin delivery (see Quintana et al. 2016 for a review on the challenges of properly administering intranasal oxytocin). Intranasal administration of oxytocin to macaque monkeys has been reported to increase concentrations of oxytocin in cerebrospinal fluid within 40 minutes (Dal Monte et al. 2014), and in humans, concentrations of vasopressin (which is structurally similar to oxytocin) in cerebrospinal fluid were shown to increase 10–80 minutes after intranasal vasopressin administration (Born et al. 2002), but direct evidence for immediate oxytocin elevations in cerebrospinal fluid in humans is still lacking (Leng and Ludwig 2015; Striepens et al. 2013). Third, the scope of the current study did not allow us to examine the dose-dependence or durability of oxytocin's effects. These questions should be

examined in future studies. Fourth, this study included only men due to Institutional Review Board concerns about off-target effects among women. Given the known sex differences in the biology of oxytocin, future studies should include women while incorporating proper precautions. Strengths of this study include the randomized, double-blind, placebo-controlled, crossover design; the use of a robust, well-established task to measure mesoaccumbal activation in humans; and the focused analyses on regions of interest, including examination of the BOLD signal time-course.

The principal findings of our study are not consistent with those from a comparable study recently reported by Nawijn and colleagues (Nawijn et al. 2016). That study examined the effects of intranasal oxytocin on a sample of trauma-exposed police officers and found enhanced responses to monetary incentives in the putamen, insula, and anterior cingulate cortex using region-of-interest analyses. They found no effects in the midbrain or NAc, and no effects that survived whole-brain correction for multiple comparisons. The monetary incentive delay task and image acquisition and processing were similar (but not identical) between that study and ours, and the trial design and procedures for drug administration were nearly identical. One potentially important difference between the two studies is in subject characteristics. Nawijn et al. studied two samples of police officers, a group with posttraumatic stress disorder and a group of trauma-exposed controls. Both men and women were included, with mean $\pm$ SD age of approximately 40 ± 10 years. Our study included only healthy men, aged 22 ± 2 years, without trauma exposure. It is possible that our relatively homogenous sample reduced extraneous sources of variance and allowed us to detect effects of oxytocin in the midbrain and PFC that were otherwise not observable. It is also possible that the effects we observed are specific to males, although this seems less likely since no interactions with sex were found in the other study (Nawijn et al. 2016). A second potentially important difference between the two studies is in the analytic methods used. The other study relied solely on SPM modeling, which requires assumptions about the shape of the hemodynamic response function, whereas we used a combination of modeling and SPM-independent analyses of the BOLD time course. Our use of the latter approach revealed prolonging of midbrain responses that was not apparent with SPM modeling. While analytic differences could explain discrepant midbrain findings, this would not account for other discrepancies, since we detected an oxytocin effect in the PFC using SPM with whole-brain correction. Finally, it appears that the oxytocin effects reported by Nawijn et al. would not survive correction for multiple comparisons. In their region-of-interest analyses, p-values ranged 0.024–0.044 before accounting for testing of 2 task conditions and 6 regions (Nawijn et al. 2016), so type I error is possible.

Our results complement and extend previous human studies of oxytocin's effects in social contexts. For instance, a study using a social form of the incentive delay task previously demonstrated that oxytocin administration is associated with heightened midbrain and NAc responses during the anticipation of social incentives [i.e. happy/angry faces, (Groppe et al. 2013)]. Similarly, Scheele and colleagues recently showed that treatment with intranasal oxytocin can enhance both midbrain and NAc responses in men viewing the face of their female partner compared to unfamiliar women (Scheele et al. 2013). It has been hypothesized that such oxytocin-induced increases in activity could be due to oxytocin's modulatory influence on dopaminergic activity in these areas and as such may be a

reflection of increased reward value or salience attributed to these salient stimuli (Groppe et al. 2013; Scheele et al. 2013). Our results extend these findings by demonstrating, for the first time, that oxytocin treatment prolongs midbrain responses during anticipation of monetary incentives. Taken together, these results indicate that oxytocin, rather than selectively influencing neural activity associated with socially salient cues, increases midbrain responses to more general incentive cues. This conclusion runs counter to the notion that oxytocin only influences social processes.

Our findings in humans are consistent with preclinical data demonstrating that oxytocin can indeed impact non-social behaviors possibly through oxytocin's influence on dopaminergic neurotransmission (Sarnyai and Kovács 2014). Extensive work has demonstrated that oxytocin treatment can lower the rate of self-administration and reduce the rewarding impact (as assessed by conditioned place preference) of drugs such as methamphetamine (Sarnyai and Kovács 2014). These effects appear to occur concomitantly with reductions in the capacity for psychostimulants to trigger dopamine release within the NAc (Qi et al. 2007). While our functional MRI methodology cannot reveal dopamine-specific neurotransmission, BOLD activity in the VTA and NAc elicited by the monetary incentive delay task has been shown to be associated with reward-related changes in dopamine release (Schott et al. 2008). Thus, it is tempting to speculate that the impact of oxytocin administration on midbrain activity that we observed reflects changes in dopaminergic activity. Future studies utilizing techniques that quantify dopaminergic neurotransmission (e.g. positron emission tomography with dopamine receptor radiotracers) would be able to answer this possibility more directly.

The BOLD signal change we observed in the midbrain occurred specifically during the late phase of the hemodynamic response. In general, because the BOLD contrast mechanism inherently filters and delays the underlying neural signal, a brief stimulus typically evokes a peak 5–6 s following the stimulus and continues to evolve over ~20 s (Friston et al. 1994; Friston et al. 1998). We found that oxytocin increased the BOLD signal during the late phase (lag ~12 s) where an undershoot is commonly observed and not during the early peak (lag ~6 s). The significance and functional implications of such a temporally delayed response is not currently known. Although we did not anticipate this phase specificity, similar pharmacologic effects have been described previously. Knutson and colleagues found that oral amphetamine caused a blunting of the early NAc BOLD signal (6-s lag) and an increase of the late NAc BOLD signal (12-s lag) in response to potential reward, but not potential loss (Knutson et al. 2004). They did not report analyses of midbrain responses. The phase-specific effects were attributed to amphetamine's ability to blunt phasic dopamine release while increasing tonic dopamine release in the NAc (Knutson et al. 2004). If we apply this interpretation to our findings, our data support a model in which oxytocin administration increases tonic activation of midbrain neurons without affecting phasic responses. In support of this possibility, recent experiments in mice have shown that oxytocin directly activates a subset of neurons in the VTA (Tang et al. 2014); how oxytocin influences dopamine dynamics in the midbrain remains unclear. Another possibility is that oxytocin affects vascular responses or neurovascular coupling, prolonging the temporal evolution of the BOLD signal. This seems unlikely as the effects we see here are condition and region specific. Our observations are certainly consistent with other models. For example,

oxytocin's effects in the periphery or in other brain regions might modulate midbrain responses indirectly.

We found some degree of valence specificity of oxytocin effects within the midbrain. We observed an increase of the VTA BOLD signal under reward conditions during the late phase of the BOLD signal response (12-s) compared to loss conditions. We also found an unexpected decrease of the late BOLD signal under the neutral condition. As such, within the VTA, oxytocin administration both increased late BOLD signal response during the anticipation of reward and reduced BOLD response during the neutral condition, effectively exaggerating the contrast between reward and neutral conditions. Our findings add to previous studies which have reported valence-specific effects of oxytocin on emotion perception and reaction times (Di Simplicio et al. 2009), attentional bias (Domes et al. 2012; Ellenbogen et al. 2011; Kim et al. 2014), eye gaze (Domes et al. 2013b), emotional recall (Kis et al. 2013), and on the neural and parasympathetic responses to emotional stimuli (Gamer and Büchel 2012; Gamer et al. 2010). The genesis of such condition-specific effects are not entirely clear. It is possible that oxytocin may alter individual's subjective experience towards the anticipatory cues. Previous MID studies indicate that self-report measures of affective reactions to anticipatory cues are correlated with mesolimbic BOLD responses during the anticipation of reward (i.e. NAc activity, Knutson 2001, Bjork 2004, Knutson 2005, Samanez-Larkin 2007). Individual differences in reward sensitivity also predict relative BOLD responses to gain over loss (Carter 2009). In order to explore these possibilities in the future, it will be important to obtain arousal and valence ratings for each of the anticipatory cues and measures of reward sensitivity before and after oxytocin administration.

Our whole-brain analyses revealed an effect of oxytocin administration on medial PFC responses selectively during anticipation of potential losses. A recent functional MRI study of autistic individuals making social judgements reported that a dorsomedial PFC region – one that overlaps with the region we observed – was activated by intranasal oxytocin (Watanabe et al. 2014) suggesting a convergence of findings on this specific region. During the monetary incentive delay task, we found that a negative BOLD response observed under the placebo condition was attenuated by oxytocin. Although the underlying physiological mechanism of the negative BOLD response is not completely understood, recent work suggests that the most likely cause is a reduction in cerebral blood flow and a lesser reduction in cerebral metabolic rate caused by active neuronal inhibition (Mullinger et al. 2014). If this mechanism is at work in our experiment, then oxytocin most likely attenuates neuronal inhibition, presumably GABAergic, in the medial PFC during anticipation of loss. This effect of oxytocin could be mediated through cortical or subcortical oxytocin receptors. Experiments in mice recently revealed that oxytocin activates a subset of medial PFC interneurons (Nakajima et al. 2014) but how these oxytocin-responsive neurons connect with local circuitry and alter cortical function remains unknown. The cortical effects we observed could also result from oxytocin's subcortical modulation of widely projecting neurochemical systems that impinge on the cortex, such as the dopaminergic and serotonergic systems (Dolen et al. 2013; Love 2014; Yoshida et al. 2009). The specificity of oxytocin's effect for the high-salience loss context was mirrored by slowing of reaction times under this condition. Increased reaction time may be a sign of increased performance monitoring,

which may engage this medial PFC region. However, we did not observe significant correlations between reaction times and BOLD responses which would be expected if these processes were causally related. Another possibility is that the potential for monetary loss was perceived as less incentivizing during oxytocin administration. This possibility could be examined in future studies by measuring subjective perceptions of incentive stimuli with and without oxytocin. In any case, the effects of oxytocin that we observed are consistent with valence-specific attenuation of prefrontal processing of incentive stimuli.

In conclusion, our results indicate that oxytocin administration influences neural responses to monetary incentive stimuli, even in the absence of a social context. These findings could be of significant interest to researchers interested in developing oxytocin-related agents for disorders characterized by dysfunctional reward processing, such as depression, schizophrenia, and substance use disorders (Kim et al. 2014; McGregor et al. 2008; Pedersen et al. 2013). While our data indicate that oxytocin administration can broadly impact neural responses in the absence of social cues, a large body of evidence suggests that oxytocin does indeed play a special role in the manifestation of social behaviors. Future studies examining the social specific and non-social specific influences of oxytocin on reward processes should reveal how and under what circumstances oxytocin modulates reward behaviors.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

This research was supported by the National Center for Research Resources (UL1 RR 024986), now the National Center for Advancing Translational Sciences (UL1 TR 000433). BJM was supported by the NIMH (K23 MH 092648). The authors thank Thomas Chenevert, PhD, for his help and advice in scanning; technologists Suzan Lowe BA, RT(R)(MR) and Sara Easler BA, RT(R)(MR) for their assistance in collecting imaging data; and Amy Skyles, PharmD, and Mona Moore, MS, RAC, at the Michigan Institute for Clinical and Health Research for providing regulatory support and guidance.

References

- Adan RA, Van Leeuwen FW, Sonnemans MA, Brouns M, Hoffman G, Verbalis JG, Burbach JP. Rat oxytocin receptor in brain, pituitary, mammary gland, and uterus: partial sequence and immunocytochemical localization. *Endocrinology*. 1995; 136:4022–4028. [PubMed: 7649111]
- Bartz JA, Zaki J, Bolger N, Ochsner KN. Social effects of oxytocin in humans: context and person matter. *Trends Cogn Sci*. 2011
- Becker, JB.; Taylor, JR. Sex differences in motivation. In: Becker, JB.; Berkley, K.; Geary, N.; Herman E, JP.; Young, EA., editors. *Sex Differences in the Brain: from Genes to Behavior*. Oxford, UK: Oxford University Press; 2008. p. 177-199.
- Behzadi Y, Restom K, Liao J, Liu TT. A component based noise correction method (CompCor) for BOLD and perfusion based fMRI. *Neuroimage*. 2007; 37:90–101. [PubMed: 17560126]
- Boccia ML, Petrusz P, Suzuki K, Marson L, Pedersen CA. Immunohistochemical localization of oxytocin receptors in human brain. *Neuroscience*. 2013; 253:155–164. [PubMed: 24012742]
- Born J, Lange T, Kern W, McGregor GP, Bickel U, Fehm HL. Sniffing neuropeptides: a transnasal approach to the human brain. *Nat Neurosci*. 2002; 5:514–516. [PubMed: 11992114]
- Carter RM, Macinnes JJ, Huettel SA, Adcock RA. Activation in the VTA and nucleus accumbens increases in anticipation of both gains and losses. *Front Behav Neurosci*. 2009; 3:21. [PubMed: 19753142]

- Cooper JC, Knutson B. Valence and salience contribute to nucleus accumbens activation. *Neuroimage*. 2008; 39:538–547. [PubMed: 17904386]
- Costa PT, McCrae RR. Normal personality assessment in clinical practice: The NEO Personality Inventory. *Psychological Assessment*. 1992; 4:5–13.
- Dal Monte O, Noble PL, Turchi J, Cummins A, Averbeck BB. CSF and blood oxytocin concentration changes following intranasal delivery in macaque. *PLoS One*. 2014; 9:e103677. [PubMed: 25133536]
- Di Simplicio M, Massey-Chase R, Cowen P, Harmer C. Oxytocin enhances processing of positive versus negative emotional information in healthy male volunteers. *J Psychopharmacol (Oxford)*. 2009; 23:241–248. [PubMed: 18801829]
- Dolen G, Darvishzadeh A, Huang KW, Malenka RC. Social reward requires coordinated activity of nucleus accumbens oxytocin and serotonin. *Nature*. 2013; 501:179–184. [PubMed: 24025838]
- Domes G, Sibold M, Schulze L, Lischke A, Herpertz SC, Heinrichs M. Intranasal oxytocin increases covert attention to positive social cues. *Psychological medicine*. 2012; 43:1747–1753. [PubMed: 23146328]
- Domes G, Sibold M, Schulze L, Lischke A, Herpertz SC, Heinrichs M. Intranasal oxytocin increases covert attention to positive social cues. *Psychol Med*. 2013a; 43:1747–1753. [PubMed: 23146328]
- Domes G, Steiner A, Porges SW, Heinrichs M. Oxytocin differentially modulates eye gaze to naturalistic social signals of happiness and anger. *Psychoneuroendocrinology*. 2013b; 38:1198–1202. [PubMed: 23117026]
- Dumais KM, Bredewold R, Mayer TE, Veenema AH. Sex differences in oxytocin receptor binding in forebrain regions: correlations with social interest in brain region- and sex- specific ways. *Horm Behav*. 2013; 64:693–701. [PubMed: 24055336]
- Eapen M, Zald DH, Gatenby JC, Ding Z, Gore JC. Using high-resolution MR imaging at 7T to evaluate the anatomy of the midbrain dopaminergic system. *AJNR Am J Neuroradiol*. 2011; 32:688–694. [PubMed: 21183619]
- Ellenbogen MA, Linnen A-M, Grumet R, Cardoso C, Joobar R. The acute effects of intranasal oxytocin on automatic and effortful attentional shifting to emotional faces. *Psychophysiology*. 2011; 49:128–137. [PubMed: 22092248]
- Friston K, Jezzard P, Turner R. Analysis of functional mri time-series. *Human Brain Mapping*. 1994; 1:153–171.
- Friston KJ, Fletcher P, Josephs O, Holmes A, Rugg MD, Turner R. Event-related fMRI: characterizing differential responses. *Neuroimage*. 1998; 7:30–40. [PubMed: 9500830]
- Gamer M, Büchel C. Oxytocin specifically enhances valence-dependent parasympathetic responses. *Psychoneuroendocrinology*. 2012; 37:87–93. [PubMed: 21641726]
- Gamer M, Zurowski B, Büchel C. Different amygdala subregions mediate valence-related and attentional effects of oxytocin in humans. *Proc Natl Acad Sci USA*. 2010; 107:9400–9405. [PubMed: 20421469]
- Gordon I, Martin C, Feldman R, Leckman JF. Oxytocin and social motivation. *Dev Cogn Neurosci*. 2011; 1:471–493. [PubMed: 21984889]
- Gregory R, Cheng H, Rupp HA, Sengelaub DR, Heiman JR. Oxytocin increases VTA activation to infant and sexual stimuli in nulliparous and postpartum women. *Horm Behav*. 2015; 69:82–88. [PubMed: 25562711]
- Groppe SE, Gossen A, Rademacher L, Hahn A, Westphal L, Gründer G, Spreckelmeyer KN. Oxytocin Influences Processing of Socially Relevant Cues in the Ventral Tegmental Area of the Human Brain. *Biol Psychiatry*. 2013; 74:172–179. [PubMed: 23419544]
- Helfinstein SM, Kirwan ML, Benson BE, Hardin MG, Pine DS, Ernst M, Fox NA. Validation of a child-friendly version of the monetary incentive delay task. *Soc Cogn Affect Neurosci*. 2013; 8:720–726. [PubMed: 22569186]
- Kemp AH, Guastella AJ. The Role of Oxytocin in Human Affect: A Novel Hypothesis. *Current Directions in Psychological Science*. 2011; 20:222–231.
- Kim Y-R, Oh S-M, Corfield F, Jeong D-W, Jang E-Y, Treasure J. Intranasal Oxytocin Lessens the Attentional Bias to Adult Negative Faces: A Double Blind within-Subject Experiment. *Psychiatry Investig*. 2014; 11:160.

- Kis A, Kemerle K, Hernádi A, Topál J. Oxytocin and social pretreatment have similar effects on processing of negative emotional faces in healthy adult males. *Front Psychol*. 2013;4. [PubMed: 23378839]
- Knutson B, Bjork JM, Fong GW, Hommer D, Mattay VS, Weinberger DR. Amphetamine Modulates Human Incentive Processing. *Neuron*. 2004; 43:261–269. [PubMed: 15260961]
- Knutson B, Fong GW, Adams CM, Varner JL, Hommer D. Dissociation of reward anticipation and outcome with event-related fMRI. *Neuroreport*. 2001; 12:3683–3687. [PubMed: 11726774]
- Krebs RM, Heipertz D, Schuetze H, Duzel E. Novelty increases the mesolimbic functional connectivity of the substantia nigra/ventral tegmental area (SN/VTa) during reward anticipation: Evidence from high-resolution fMRI. *Neuroimage*. 2011; 58:647–655. [PubMed: 21723396]
- Lee MR, Glassman M, King-Casas B, Kelly DL, Stein EA, Schroeder J, Salmeron BJ. Complexity of oxytocin effects in a chronic cocaine dependent population. *Eur Neuropsychopharmacol*. 2014; 24:1483–1491. [PubMed: 25044050]
- Leng G, Ludwig M. Intranasal Oxytocin: Myths and Delusions. *Biol Psychiatry*. 2015
- Loup F, Tribollet E, Dubois-Dauphin M, Dreifuss JJ. Localization of high-affinity binding sites for oxytocin and vasopressin in the human brain. An autoradiographic study. *Brain Res*. 1991; 555:220–232. [PubMed: 1657300]
- Love TM. Oxytocin, motivation and the role of dopamine. *Pharmacol Biochem Behav*. 2014; 119:49–60. [PubMed: 23850525]
- Marsh AA, Yu HH, Pine DS, Blair RJR. Oxytocin improves specific recognition of positive facial expressions. *Psychopharmacology*. 2010; 209:225–232. [PubMed: 20186397]
- McGregor IS, Callaghan PD, Hunt GE. From ultrasocial to antisocial: a role for oxytocin in the acute reinforcing effects and long-term adverse consequences of drug use? *Br J Pharmacol*. 2008; 154:358–368. [PubMed: 18475254]
- Melis MR, Melis T, Cocco C, Succu S, Sanna F, Pillolla G, Boi A, Ferri GL, Argiolas A. Oxytocin injected into the ventral tegmental area induces penile erection and increases extracellular dopamine in the nucleus accumbens and paraventricular nucleus of the hypothalamus of male rats. *Eur J Neurosci*. 2007; 26:1026–1035. [PubMed: 17672853]
- Melis MR, Succu S, Sanna F, Boi A, Argiolas A. Oxytocin injected into the ventral subiculum or the posteromedial cortical nucleus of the amygdala induces penile erection and increases extracellular dopamine levels in the nucleus accumbens of male rats. *Eur J Neurosci*. 2009; 30:1349–1357. [PubMed: 19769589]
- Mullinger KJ, Mayhew SD, Bagshaw AP, Bowtell R, Francis ST. Evidence that the negative BOLD response is neuronal in origin: a simultaneous EEG-BOLD-CBF study in humans. *Neuroimage*. 2014; 94:263–274. [PubMed: 24632092]
- Nakajima M, Gorlich A, Heintz N. Oxytocin modulates female sociosexual behavior through a specific class of prefrontal cortical interneurons. *Cell*. 2014; 159:295–305. [PubMed: 25303526]
- Nawijn L, van Zuiden M, Koch SB, Frijling JL, Veltman DJ, Olf M. Intranasal oxytocin enhances neural processing of monetary reward and loss in post-traumatic stress disorder and traumatized controls. *Psychoneuroendocrinology*. 2016; 66:228–237. [PubMed: 26851698]
- Pedersen CA, Smedley KL, Leserman J, Jarskog LF, Rau SW, Kampov-Polevoi A, Casey RL, Fender T, Garbutt JC. Intranasal oxytocin blocks alcohol withdrawal in human subjects. *Alcohol Clin Exp Res*. 2013; 37:484–489. [PubMed: 23025690]
- Qi J, Yang J-Y, Song M, Li Y, Wang F, Wu C-F. Inhibition by oxytocin of methamphetamine-induced hyperactivity related to dopamine turnover in the mesolimbic region in mice. *Naunyn-Schmied Arch Pharmacol*. 2007; 376:441–448.
- Quintana DS, Guastella AJ, Westlye LT, Andreassen OA. The promise and pitfalls of intranasally administering psychopharmacological agents for the treatment of psychiatric disorders. *Mol Psychiatry*. 2016; 21:29–38. [PubMed: 26552590]
- Riem MME, Bakermans-Kranenburg MJ, Pieper S, Tops M, Boksem MAS, Vermeiren RRJM, van Ijzendoorn MH, Rombouts SAR. Oxytocin Modulates Amygdala, Insula, and Inferior Frontal Gyrus Responses to Infant Crying: A Randomized Controlled Trial. *Biol Psychiatry*. 2011a; 70:291–297. [PubMed: 21470595]

- Riem MME, van Ijzendoorn MH, Tops M, Boksem MAS, Rombouts SARB, Bakermans-Kranenburg MJ. No Laughing Matter: Intranasal Oxytocin Administration Changes Functional Brain Connectivity during Exposure to Infant Laughter. *Neuropsychopharmacology*. 2011b; 37:1257–1266. [PubMed: 22189289]
- Rilling JK, DeMarco AC, Hackett PD, Chen X, Gautam P, Stair S, Haroon E, Thompson R, Ditzen B, Patel R, Pagnoni G. Sex differences in the neural and behavioral response to intranasal oxytocin and vasopressin during human social interaction. *Psychoneuroendocrinology*. 2014; 39:237–248. [PubMed: 24157401]
- Rimmele U, Hediger K, Heinrichs M, Klaver P. Oxytocin Makes a Face in Memory Familiar. *J Neurosci*. 2009; 29:38–42. [PubMed: 19129382]
- Sarnyai Z, Kovács GL. Oxytocin in learning and addiction: From early discoveries to the present. *Pharmacol Biochem Behav*. 2014; 119:3–9. [PubMed: 24280016]
- Scheele D, Kendrick KM, Khouiri C, Kretzer E, Schläpfer TE, Stoffel-Wagner B, Güntürkün O, Maier W, Hurlmann R. An Oxytocin-Induced Facilitation of Neural and Emotional Responses to Social Touch Correlates Inversely with Autism Traits. *Neuropsychopharmacology*. 2014; 39:2078–2085. [PubMed: 24694924]
- Scheele D, Wille A, Kendrick KM, Stoffel-Wagner B, Becker B, Gunturkun O, Maier W, Hurlmann R. Oxytocin enhances brain reward system responses in men viewing the face of their female partner. *Proc Natl Acad Sci USA*. 2013; 110:20308–20313. [PubMed: 24277856]
- Schott BH, Minuzzi L, Krebs RM, Elmenhorst D, Lang M, Winz OH, Seidenbecher CI, Coenen HH, Heinze HJ, Zilles K, Duzel E, Bauer A. Mesolimbic Functional Magnetic Resonance Imaging Activations during Reward Anticipation Correlate with Reward-Related Ventral Striatal Dopamine Release. *J Neurosci*. 2008; 28:14311–14319. [PubMed: 19109512]
- Sheehan DV, Lecrubier Y, Sheehan KH, Amorim P, Janavs J, Weiller E, Hergueta T, Baker R, Dunbar GC. The Mini-International Neuropsychiatric Interview (M.I.N.I.): the development and validation of a structured diagnostic psychiatric interview for DSM-IV and ICD-10. *J Clin Psychiatry* 59 Suppl. 1998; 20:22, 33. quiz 34-57.
- Spielberger, CD.; Gorssuch, RL.; Lushene, PR.; Vagg, PR.; Jacobs, GA. Manual for the State-Trait Anxiety Inventory. Consulting Psychologists Press, Inc; 1983.
- Striepen N, Kendrick KM, Hanking V, Landgraf R, Wullner U, Maier W, Hurlmann R. Elevated cerebrospinal fluid and blood concentrations of oxytocin following its intranasal administration in humans. *Sci Rep*. 2013; 3:3440. [PubMed: 24310737]
- Tang Y, Chen Z, Tao H, Li C, Zhang X, Tang A, Liu Y. Oxytocin activation of neurons in ventral tegmental area and interfascicular nucleus of mouse midbrain. *Neuropharmacology*. 2014; 77:277–284. [PubMed: 24148809]
- Watanabe T, Abe O, Kuwabara H, Yahata N, Takano Y, Iwashiro N, Natsubori T, Aoki Y, Takao H, Kawakubo Y, Kamio Y, Kato N, Miyashita Y, Kasai K, Yamasue H. Mitigation of sociocommunicational deficits of autism through oxytocin-induced recovery of medial prefrontal activity: a randomized trial. *JAMA Psychiatry*. 2014; 71:166–175. [PubMed: 24352377]
- Watson, D.; Clark, LA. PANAS-X Manual for the Positive and Negative Affect Schedule -Expanded Form. University of Iowa; 1994.
- Wittfoth-Schardt D, Grunding J, Wittfoth M, Lanfermann H, Heinrichs M, Domes G, Buchheim A, Gundel H, Waller C. Oxytocin Modulates Neural Reactivity to Children's Faces as a Function of Social Salience. *Neuropsychopharmacology*. 2012; 37:1799–1807. [PubMed: 22510724]
- Yoshida M, Takayanagi Y, Inoue K, Kimura T, Young LJ, Onaka T, Nishimori K. Evidence that oxytocin exerts anxiolytic effects via oxytocin receptor expressed in serotonergic neurons in mice. *J Neurosci*. 2009; 29:2259–2271. [PubMed: 19228979]

Fig 1. Design of the monetary incentive delay task

Participants were presented with a cue (i.e., stimulus) at the beginning of each trial indicating the type of trial: high-salience win (W?), low-salience win (W), high-salience loss (L?), low-salience loss (L), or neutral (N). The cue was followed by crosshairs during a variable delay period, and then the brief appearance of a target (solid white square). Participants hit the target by pressing a button before the target disappeared. At the end of the trial, the participant received feedback about the outcome of the trial (rectangle at right). Trials were separated by jittered inter-trial intervals (ITI) of 2–6 seconds.

Fig 2. Anatomical locations of volumes of interest

Volumes of interest in left and right nucleus accumbens and midbrain (ventral tegmental area and substantia nigra) are shown in color (blue, green, and red, respectively) overlaid on a Montreal Neurological Institute T1-weighted template image. Panels show coronal, parasagittal, and horizontal sections (upper left, upper right, and lower left, respectively).

Fig 3. Effects of oxytocin on hemodynamic response in the midbrain

The blood oxygenation level dependent (BOLD) signal in the midbrain (ventral tegmental area and substantia nigra) is shown versus time after presentation of the cue (beginning of the trial). Thick lines represent the mean across 18 subjects and thin lines indicate ± 1 standard error. Black dotted lines represent the placebo condition and red solid lines indicate the oxytocin condition. Four contrasts are shown: (A) low-salience reward - neutral, (B) high-salience reward - neutral, (C) low-salience loss - neutral, (D) high-salience loss - neutral.

Fig 4. No significant effects of oxytocin in nucleus accumbens

Blood oxygenation level dependent (BOLD) response in left nucleus accumbens (NAc) is shown using the conventions of Figure 3. Similar results were obtained from the right NAc (data not shown).

Fig 5. Effects of oxytocin on hemodynamic response of medial prefrontal cortex

Statistical parametric map shows regions in which the modeled response is greater during the oxytocin scan relative to the placebo scan during anticipation of monetary loss (high-salience loss - neutral, paired t test, $n = 18$). Heat maps represent uncorrected, two-tailed p values, thresholded at $p < 0.01$. The significant cluster in medial prefrontal cortex (PFC) is shown in (A) coronal, (B) sagittal, and (C) horizontal sections overlaying an MNI standardized brain. (D) Blood oxygen level dependent (BOLD) signal time course was extracted from the significant medial PFC cluster for oxytocin and placebo conditions, and plotted using the conventions of Figure 3.

Fig 6. Oxytocin-related change in midbrain response is associated with oxytocin-related change in positive emotional state

(A) The BOLD-signal difference between oxytocin and placebo scans (Δ BOLD signal) is shown as a paired t statistic versus time after cue presentation (all task conditions relative to neutral). The dotted line represents the critical value of t ($p = 0.05$, two-tailed). The effect of oxytocin is significant at 12 s. (B) Oxytocin-related change in midbrain response is correlated with oxytocin-related change in emotional state. The Pearson correlation r between change in the Positive and Negative Affect Schedule-positive scale (Δ PANAS-positive) and Δ BOLD signal is shown versus time after cue presentation. The dotted line

represents the critical value of r ($p = 0.05$, two-tailed). Emotional change is significantly correlated with midbrain response at 6 s (blue circle) and 12 s (red circle). (C, D) Scatterplots of BOLD signal versus PANAS-positive are shown for 6-s and 12-s correlations, respectively, that are shown in (B).

Table 1

Effects of oxytocin on behavior, physiology, and emotional state

	Oxytocin Mean (SD)	Placebo Mean (SD)	Hedges g	Paired t test t	p
Performance during monetary incentive delay task					
Hit Rate (%)					
Low-salience Reward	56.8 (13.7)	55.7 (13.1)	0.101	0.44	0.67
High-salience Reward	70.2 (10.9)	72.9 (10.0)	-0.218	-0.94	0.36
Low-salience Loss	47.5 (13.1)	48.1 (11.2)	-0.042	-0.18	0.86
High-salience Loss	72.2 (12.1)	76.6 (7.7)	-0.370	-1.61	0.13
Neutral	49.8 (18.0)	50.3 (14.1)	-0.037	-0.16	0.88
Overall	59.3 (8.0)	60.8 (6.3)	-0.267	-1.16	0.26
Reaction Time (ms)					
Low-salience Reward	278 (57)	269 (37)	0.237	1.03	0.32
High-salience Reward	248 (42)	234 (42)	0.388	1.69	0.11
Low-salience Loss	296 (68)	283 (57)	0.357	1.55	0.14
High-salience Loss	252 (42)	227 (44)	0.508	2.21	0.04 *
Neutral	303 (67)	295 (65)	0.124	0.54	0.60
Overall	275 (49)	262 (34)	0.564	2.45	0.03 *
Physiological measures during scan					
Heart rate (per minute)	63.3 (12.0)	62.2 (10.1)	0.224	0.92	0.37
Respiratory rate (per minute)	15.6 (3.3)	16.2 (2.5)	-0.147	-0.60	0.56
Emotional state measures					
PANAS Composite	13.4 (6.4)	13.7 (8.9)	-0.027	-0.12	0.91
Positive Scale	25.6 (6.3)	25.5 (7.9)	0.007	0.03	0.97
Negative Scale	12.1 (2.6)	11.8 (4.2)	0.076	0.33	0.75
STAI State	35.0 (6.4)	33.2 (6.3)	0.272	1.18	0.26
BFNE Total	30.9 (7.2)	30.6 (7.2)	0.061	0.27	0.79

* p < 0.05, uncorrected;

PANAS, Positive and Negative Affect Schedule; STAI, State-Trait Anxiety Inventory; BFNIE, Brief Fear of Negative Evaluation scale; $df = 17$ for all t tests, except $df = 15$ for heart and respiratory rates

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Table 2

Analysis of variance for a linear mixed model of midbrain hemodynamic response time-course after treatment with oxytocin or placebo (6–12 second lag)

	χ^2	df	p *
4-level model (neutral subtracted)			
Time	19.1	1	1.2×10^{-5}
Treatment	0.34	1	0.56
Condition	74.3	3	5.2×10^{-16}
Time $\times$ Treatment	8.35	1	0.0038
Treatment $\times$ Condition	4.60	3	0.20
Time $\times$ Condition	21.9	3	6.8×10^{-5}
Time $\times$ Treatment $\times$ Condition	1.72	3	0.63
5-level model (including neutral)			
Time	35.2	1	3.0×10^{-9}
Treatment	0.825	1	0.36
Condition	102	4	$<2.2 \times 10^{-16}$
Time $\times$ Treatment	6.32	1	0.012
Treatment $\times$ Condition	16.8	4	0.0021
Time $\times$ Condition	37.9	4	1.2×10^{-7}
Time $\times$ Treatment $\times$ Condition	7.61	4	0.11

* Type II Wald chi-squared tests

4-level model: Task condition is modeled as a factor with 4 levels: low-salience reward – neutral, high-salience reward – neutral, low-salience loss – neutral, and high-salience loss – neutral.

5-level model: Task condition is modeled as a factor with 5 levels: low-salience reward, high-salience reward, low-salience loss, high-salience loss, and neutral.

Table 3
SPM8 whole-brain analysis of oxytocin effects on hemodynamic response to high-salience loss

Cluster location	Cluster-level statistics					MNI peak coordinates (mm)
	p (FWE)	p (FDR)	p (unc)	extent (voxels)	peak t	
Medial prefrontal	1×10^{-7}	2×10^{-7}	7×10^{-9}	768	7.48	-10 38 48
					6.51	-10 26 58
					5.88	0 46 36
Left dorsolateral prefrontal	0.10	0.06	0.01	105	5.79	-50 18 42
					3.91	-50 16 32
Left lateral prefrontal	0.05	0.05	0.004	126	5.01	-50 26 16
					4.21	-48 32 26

SPM8 paired t test, n = 18 subjects, not corrected for testing 4 contrasts; threshold p = 0.001 (one-sided, uncorrected); threshold cluster size = 100 voxels; FWE, family-wise error correction; FDR, false discovery rate correction; unc, uncorrected; MNI, Montreal Neurological Institute